CLINICAL LARGE INTESTINAL COCCIDIOSIS IN CAMELS (*CAMELUS DROMEDARIUS*) IN THE UNITED ARAB EMIRATES: DESCRIPTION OF LESIONS, ENDOGENOUS STAGES, AND REDESCRIPTION OF *ISOSPORA ORLOVI*, TSYGANKOV, 1950 OOCYSTS

Joerg Kinne, Mansoor Ali*, Ulrich Wernery, and J. P. Dubey†

Central Veterinary Research Laboratory, P.O. Box 597, Dubai, United Arab Emirates. e-mail: jdubey@anri.barc.usda.gov

ABSTRACT: Between January and March 2001, eight 4- to 8-wk-old camels (*Camelus dromedarius*) from 2 farms from Dubai area of the United Arab Emirates were submitted for necropsy examination. The camels had diarrhea of 2–5 days duration. Grossly, a severe diphtheroid-to-hemorrhagic colitis was seen in all animals. Gamonts, unsporulated oocysts, sporulating oocysts, and fully sporulated oocysts were present in the intestinal epithelium and the lamina propria. Fully sporulated oocysts contained 2 sporocysts and 4 sporozoites in each sporocyst. Oocysts from fecal samples resembled oocysts of *Isospora orlovi*. This is the first report of an isosporan parasite associated with hemorrhagic enteritis in the large intestine of any animal.

Five species of *Eimeria*, *Eimeria bactriani* Levine and Ivens, 1970, *Eimeria cameli* Henry and Masson, 1932; Reichnow, 1952, *Eimeria dromedarii* Yakimoff and Matschoulsky, 1939, *Eimeria pellérdyi* Prasad, 1960 emend. Pellérdy, 1965, and *Eimeria rajasthani* Dubey and Pande, 1963, and 1 species of *Isospora*, *Isospora orlovi* Tsygankov, 1950, have been reported from feces of camels (reviewed by Dubey and Pande, 1964; Levine and Ivens, 1970). The validity of some of these coccidian species is in doubt because the original descriptions were not complete and because their life cycles have not been completed.

Although there are several reports of diarrhea associated with *Eimeria* spp. infections in camels, most of them were based only on finding oocysts in feces (Iyer et al., 1968; Chineme, 1980; Yagoub, 1989). Henry and Masson (1932), Hussein et al. (1987), Iyer et al. (1968), and Kinne and Wernery (1997) reported schizonts and gamonts of *Eimeria* spp. in the small intestine of camels. There is 1 report of *I. orlovi*-like oocysts, possibly associated with clinical coccidiosis in a 6-mo-old camel from India (Raisinghani et al., 1987).

The objective of the present paper is to document clinical coccidiosis in camels associated with an *I. orlovi*-like parasite.

MATERIALS AND METHODS

Between January and March 2001, an outbreak of a camel calf disease occurred in the Dubai area. Calves on 2 camel farms had diarrhea lasting 2–5 days, with no response to treatment with an anticoccidial (Baycox, 2.5% for 5 days, 0.2 ml/kg body weight). The mother camels, which were kept in large pens, received hay ad libitum and small amounts of grain pellets. When the calves became sick, they were kept in smaller pens together with their mothers. Adult camels were apparently normal.

A total of 22 camel calf carcasses was submitted for necropsy examination. All necropsies were performed within 1–5 hr after death. Pieces of intestines, liver, spleen, and lymph nodes were taken for microbiological investigations using routine methods. The intestinal samples were also tested for the growth of anaerobes.

For histopathology, small pieces of all organs, including compartment 1 and stomach, as well as muscles and lymph nodes were fixed for 24

hr in buffered neutral 10% formalin. Paraffin-embedded 5-µm-thick sections were stained with hematoxylin and eosin, or periodic acid Schiff hematoxylin.

Oocysts were collected from feces of each camel by flotation in sucrose solution, suspended in 2.5% potassium dichromate, and examined microscopically. Oocysts from camels and sections of intestines from 3 camels were sent to the Parasite Biology, Epidemiology and Systematics Laboratory, Beltsville, Maryland, for further evaluation. Photographs of oocysts are from feces removed from colon of camel no. 269-01.

RESULTS

Lesions

Severe pseudomembraneous-to-hemorrhagic colitis was found in 8 calves from 2 farms (Fig. 1A). Microscopically, the large intestinal mucosa was hemorrhagic and infiltrated by leukocytes, including neutrophils, eosinophils, and macrophages (Fig. 1A, C). Microbiological investigations, including anaerobic cultures, did not detect any pathogenic bacteria.

Endogenous stages

Numerous coccidian stages (gamonts and oocysts) were located in the colon (Fig. 2) but not in the small intestine. Under the low power of the microscope, masses of coccidian stages were visible, often as a raised area (Fig. 1B). Only gamonts and oocysts were identified with certainty. All stages of gametogony and sporogony were present (Fig. 2). Multinucleated microgamonts-to-free microgametes were present among macrogamonts (Fig. 2A, B). Uninucleate macrogamonts-to-fully sporulated oocysts were seen. Oocysts contained 2 sporocysts, each with 4 sporozoites (Figs. 2G, 3F). Sporozoites in sections varied in size (Fig. 2G–I). In appropriately cut sections, there was a central nucleus and 2 eosinophilic bodies on either side of the sporozoite nucleus (Fig. 2H). One group of free zoites was also present (Fig. 2F). Sporozoites were 6–12 µm in section (Fig. 2G, H).

Description of oocysts

Oocysts in different stages of sporogony were present in colon contents (Fig. 3). Immature unsporulated oocysts were ovoid and contained a granulated mass (sporont) occupying the entire oocyst. These immature unsporulated oocysts (Fig. 3A–C) were $22-23\times 17-23~\mu m$ in size (n = 10). Unsporulated oocysts that appeared ready for sporulation were ovoid and had a central contracted sporont (Fig. 3D). These oocysts were $27-33\times 20-27~\mu m$ in size (n = 10).

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^{*}Camel Reproduction Laboratory, P.O. Box 9229, Nakhlee, Dubai, United Arab Emirates.

[†]To whom correspondence should be addressed.United States Department of Agriculture, Agricultural Research Service, Animal and Natural Resources Institute, Parasite Biology, Epidemiology and Systematics Laboratory, BARC-East, Bldg. 1001, Beltsville, Maryland 20705-2350.

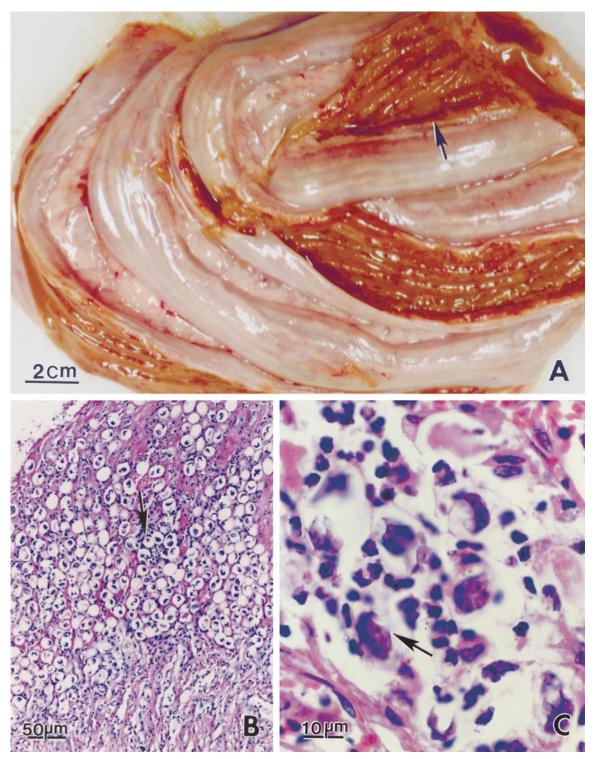


FIGURE 1. Lesions and coccidia in colon of camels. (A) Hemorrhages in colon mucosa (arrows). Unstained. (B) Masses of coccidian oocysts (arrow) in a raised area. Hematoxylin and eosin stain. (C) Focal enteritis around degenerating coccidia. Note the necrosis and infiltration by neutrophils. Hematoxylin and eosin stain.

Sporulated oocysts varied in shape, depending on the arrangement of sporocysts. Sporulated oocysts contained 2 sporocysts that were remarkably constant in size (Fig. 3E, F). Sporocysts freed from oocysts were $20 \times 15 \mu m$ in size (n = 10). In some sporocysts, all 4 sporozoites were visualized. Sporocysts

rozoites varied in shape and size, presumably depending on their viability (Fig. 3F–H). Elongated sporozoites were bananashaped and measured $12-14\times3-4~\mu m$ (n = 20) in size (Fig. 3F). Smaller, stubby sporozoites had rounded ends and were much smaller than elongated sporozoites (Fig. 3G, H). Occa-

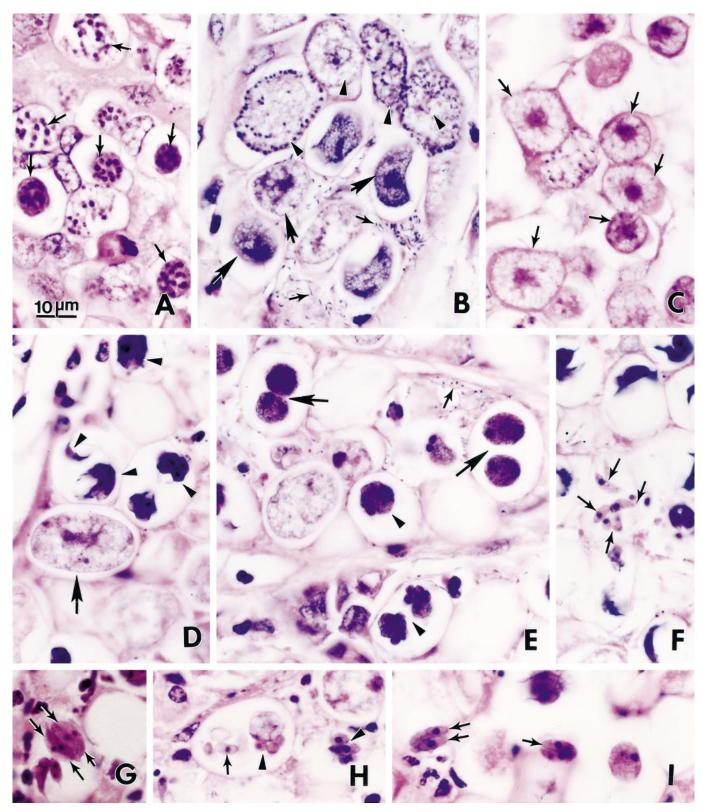


FIGURE 2. Coccidian stages in sections of colon of camels. Hematoxylin and eosin stain. Bar applies to all figures. (A) Immature multinucleated microgaments (arrows). (B) Microgaments (arrowheads), free microgametes (small arrows), and macrogaments and unsporulated oocysts (large arrows). (C) Uninucleate macrogaments (arrows). (D) Unsporulated oocyst (large arrow) and sporulating oocysts (arrowheads). (E) Sporulating oocysts with 2 sporocysts (large arrows), sporocysts with developing sporozoites (arrowhead), and free microgametes (small arrow). (F) A group of free zoites (arrows). (G) Longitudinally cut sporozoites (arrows) in a sporocyst. (H) Sporozoites (arrowheads) and 1 with a central nucleus (arrow). (I) Sporocysts with sporozoites (arrows).

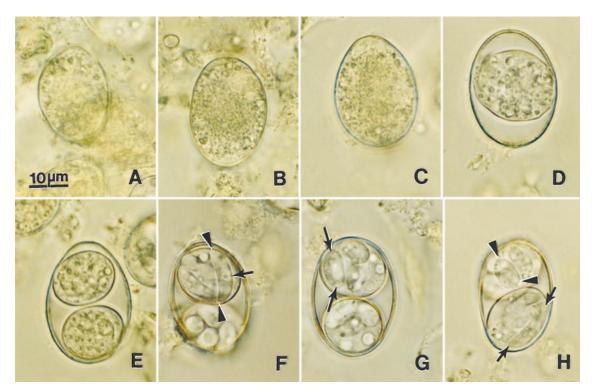


FIGURE 3. *Isospora orlovi* oocysts from colon contents of camel calf no. 269-01. The oocysts were floated and suspended in 2.5% potassium dichromate. Hanging drop preparations of oocysts sealed between a glass slide and a coverslip were used to photograph oocysts suspended in potassium dichromate. Unstained. Bar applies to all figures. (A–C) Immature unsportlated oocysts with sporont occupying the entire oocyst. (D) Unsportlated oocyst with a contracted sporont. (E) Oocyst with 2 sporoblasts. (F) Sportlated oocyst with 2 sporocysts. Note a central nucleus (arrow) in 1 of the longitudinally oriented sporozoites (arrowheads). (G) Oocyst with ovoid sporozoites (arrows). (H) Oocyst with elongated (arrows) and stubby sporozoites (arrowheads).

sionally, both elongated and ovoid sporozoites were seen in the same oocyst but in separate sporocysts (Fig. 3H). A micropyle, polar granule, oocyst residuum, and Stieda bodies were absent.

Specimens deposited

A hematoxylin and eosin-stained section of colon of camel has been deposited in the U.S. National Parasite Collection, Beltsville, Maryland, as USNPC no. 91768.

DISCUSSION

The most unusual aspects of the present study of coccidiosis in camel are the association of an isosporan parasite with clinical colitis and the association of gamonts and oocysts with lesions, in the absence of recognizable merozoites and schizonts. There is no report of any isosporan causing severe coccidiosis of large intestine in any animal species. The coccidial lesion in camels is similar to clinical coccidiosis of large intestine of cattle associated with *Eimeria bovis* and *Eimeria zuernii* (Hammond, 1973). The only isosporan parasite of clinical significance in livestock is *Isospora suis* in pigs. *Isospora suis* causes enteritis in baby pigs, mainly affecting the small intestine (Lindsay et al., 1980; Stuart et al., 1980).

Another unusual feature of the present report is the identification of sporogony in the large intestine. The only coccidian parasite that sporulates in the gut of mammals is *Sarcocystis* spp., but sporogony takes place in the lamina propria of carnivores and is not associated with clinical signs (Dubey et al.,

1989). The parasite of camel colon in the present study is probably not *Sarcocystis* because oocysts have relatively thick oocyst walls; therefore, free sporocysts are not found in feces.

Whether the *Isospora* sp. in the camel intestine sporulated in situ could not be determined because tissues were not fixed until 1–5 hr after death, and tissues had been fixed in formalin for at least 5 hr before processing for histologic sections. However, it is likely that the oocysts sporulated in situ before death because oxygen is needed for sporulation of coccidian oocysts. The type of host cell parasitized and the location of the parasite development were not identified because most of the epithelium had been destroyed or sloughed.

Oocysts of the parasite in camel feces are *Isospora* spp. In feces of camel no. 1 (269-01), unsporulated and sporulated oocysts were present. Unsporulated oocysts with an irregular shape and sporont occupying the entire oocyst were considered to have sloughed from the epithelium before the sporont had condensed into a single central mass. Oocysts containing the sporont occupying the entire oocysts usually do not sporulate.

Isospora orlovi was named by Tsygankov (1950) for oocysts in ten of nineteen 10- to 35-day-old camels from Alma-Ata, Kazakhstan, of the former USSR. A full description of oocysts was promised by Tsygankov but not published. Only drawings were published, and they are the same as those given by Levine and Ivens (1970). Isospora orlovi oocysts were $27-35 \times 15-20 \mu m$ in size, without a micropyle, residuum, or polar granule. Sporocysts were $15-20 \times 13-17 \mu m$ in size and without a

Stieda body. Sporozoites were elongate, ellipsoidal, $7-10 \times 4-6 \mu m$ in size (Tsygankov, 1950; Levine and Ivens, 1970).

Raisinghani et al. (1987) reported *I. orlovi*-like oocysts in a 6-mo-old *Camelus dromedarius* from India. The oocysts were $25–35\times17–21~\mu m$, sporocysts were $13–15\times9–11~\mu m$, and sporozoites were $6–8\times4–5~\mu m$ in size (Raisinghani et al., 1987).

In the present study, oocysts from feces were $27-33 \times 20-$ 27 μ m, sporocysts were 20 \times 15 μ m, and sporozoites were 12– $14 \times 3-4$ µm in size. Thus, there is a general agreement about the size of oocysts and sporocysts but not of sporozoites. The differences in sporozoite size in these 3 studies are either related to whether sporozoites were live or dead or to different species of Isospora. Because Isospora spp. are not parasites of domestic herbivores, Péllerdy (1965) thought I. orlovi might be an avian parasite that camels might have accidentally ingested from avian feces. The finding of endogenous stages of Isospora in camels in the present study leaves no doubt that the Isospora sp. is a true parasite of camels. Moreover, the absence of a Stieda body in sporocysts also suggests that this parasite is not related to an avian Isospora. The absence of schizonts is puzzling, but schizogony might have occurred before animals developed clinical signs. One group of free zoites was seen among oocysts, but the stage of the parasite was not identified.

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